p7.5tk

7.5 k promoter

Not I   Apa I

tk coding sequence

GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACCGGCCGC CATG GGC CCG GCC GCC AAC GGC GGA

Met Gly Pro Ala Ala Asn Gly Gly

pE/Ltk

synthetic E/L promoter

Not I   Apa I

tk coding sequence

GGCCAAAAATTGAAATTTATTTTTTTTTTTTTGGAATATAAAGCGGCCGC CATG GGC CCG GCC GCC AAC GGC GGA

Met Gly Pro Ala Ala Asn Gly Gly

FIGURE 1

1. p7.5tk

7.5K PROMOTER   NOTI   APAI

5'- GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACGCGGCCCGCCATGGGCCCGGCC -3'

2. p7.5/ATG0/tk

7.5K PROMOTER   NOTI   BAMHI SMAI   PSTI

5'- GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACGCGGCCGCGGATCCCCGGGGCTGCAGGAA

SALI   TRANSLATION   TRANSCRIPTION

TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGGGCCTAACTAACTAATTTGTTTTTGT   STOP CODONS   STOP SIGNAL

APAI

GGGCCCGGCC -3'

FIGURE 2

3. p7.5/ATG1/tk

5'-GGCCAAAATTGAAAAACTAGATCTATTTATTGCACGCGGCCGCCATGGTGGATCCCCGGGCTGCAGGAA

     7.5K PROMOTER   NOTI   START CODON   BAMHI   SMAI   PSTI

TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGGGGGCCTAACTAACTAATTTGTTTTTTGT

     SALI   STOP CODONS   TRANSLATION STOP SIGNAL   TRANSCRIPTION

APAI

GGGCCCGGCC -3'

FIGURE 2 (CONT)

4. p7.5/ATG2/tk

          7.5K PROMOTER   NOTI   START CODON   BAMHI SMAI   PSTI

                                                           START   TRANSLATION

5'- GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACGGGGCCGCCATGAGTGGATCCCCGGGGCTGCAGGAA

               SALI   TRANSLATION   TRANSCRIPTION

                                      STOP CODONS   STOP SIGNAL
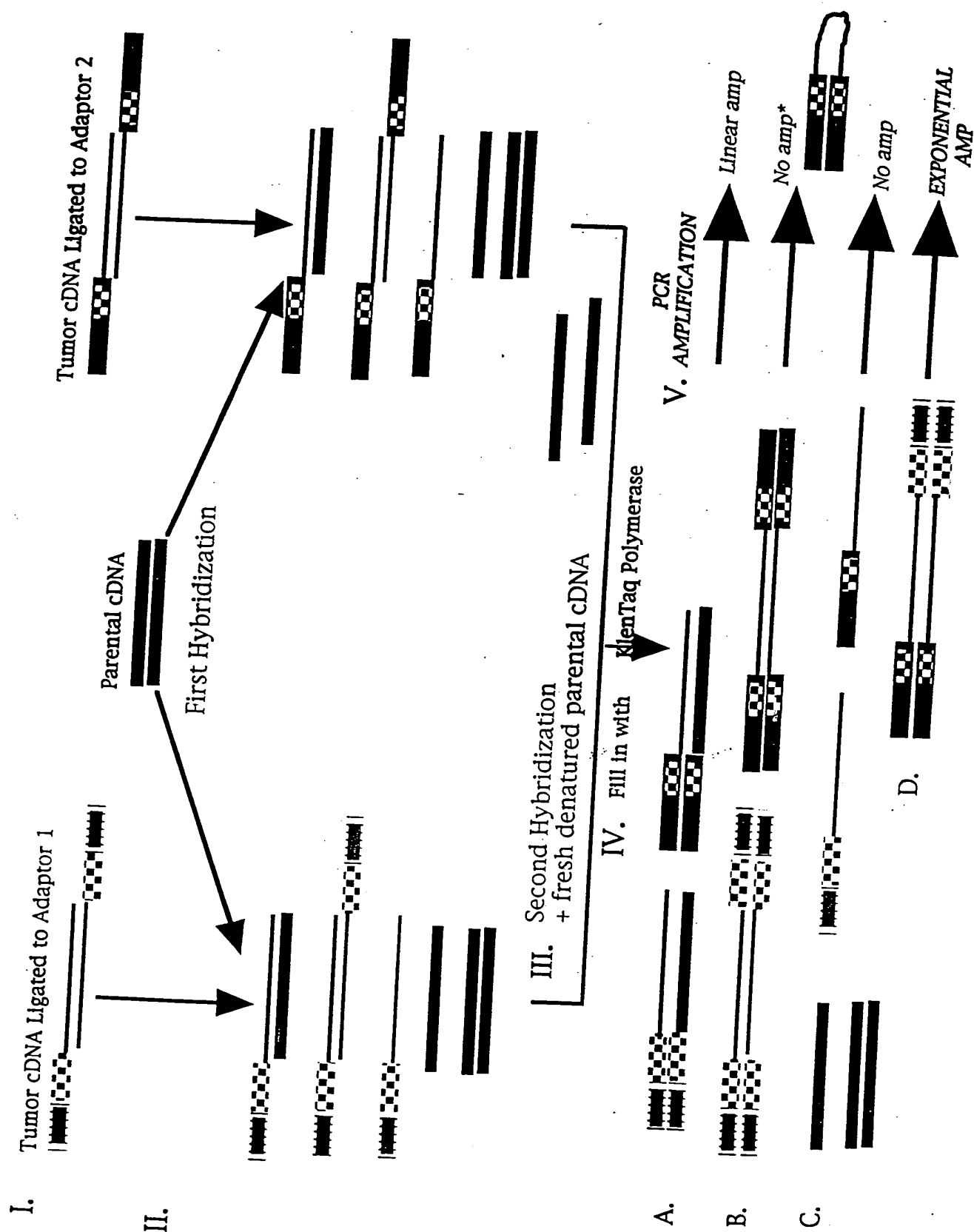
TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGGCCTAACTAACTAATTTGTTTTTGT

APAI

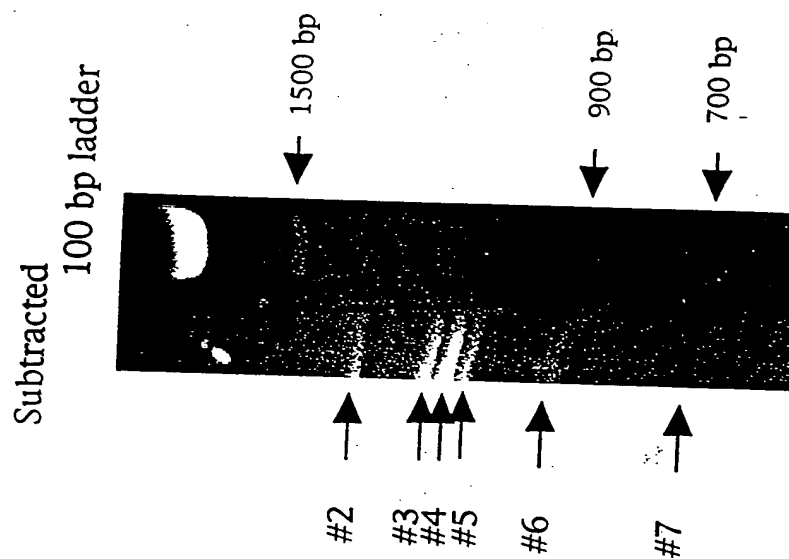GGGCCCGGCC -3'

FIGURE 2 (CONT)

5. p7.5/ATG3/tk

7.5K PROMOTER   START

NOTI CODON BAMHI SMAi PSTI

5'- GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACGGCGGCCATGACGTGGATCCCCGGGGCTGCAGGAA

SALI TRANSLATION TRANSCRIPTION

STOP CODONS STOP SIGNAL

TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGGCCTAACTAACTAATTTTGTTTTTGT
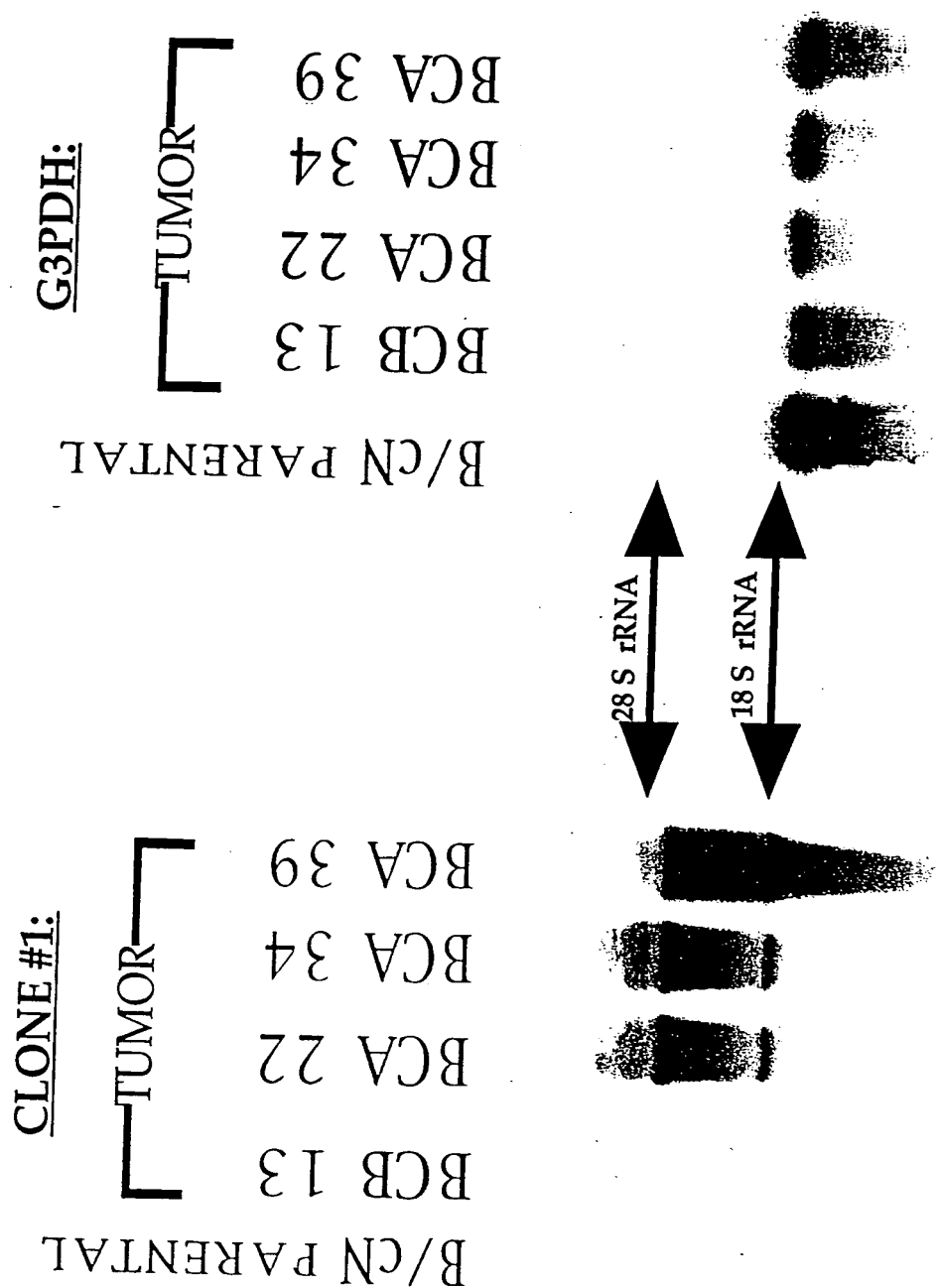
APAI

GGGCCCGGGCC -3'

FIGURE 2 (CONT)

FIGURE 3

FIGURE 4

G3PDH:

TUMOR
BCA 39
BCA 34
BCA 22
BCB 13

B/cN PARENTAL

28 S rRNA

18 S rRNA

CLONE #1:

TUMOR
BCA 39
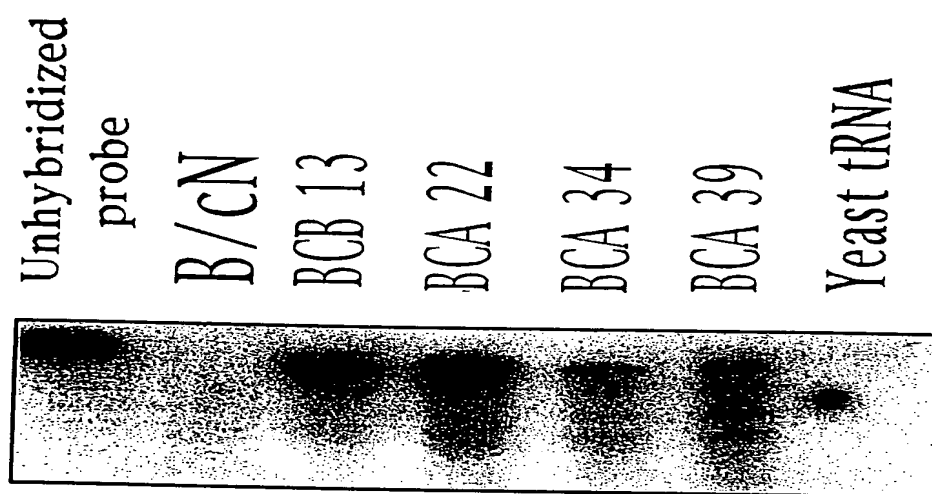BCA 34
BCA 22
BCB 13

B/cN PARENTAL
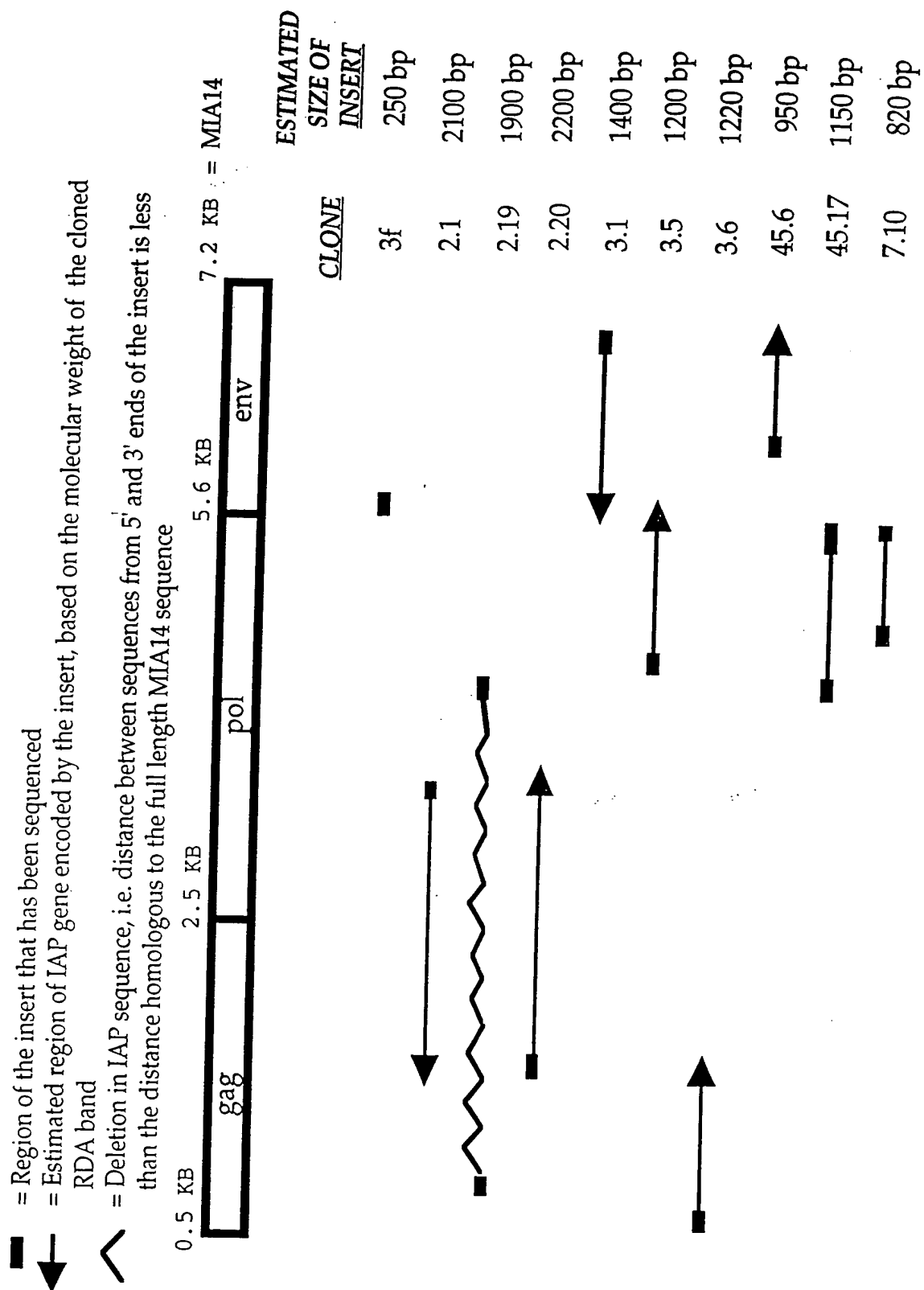
FIGURE 5

FIGURE 6

a b c d e
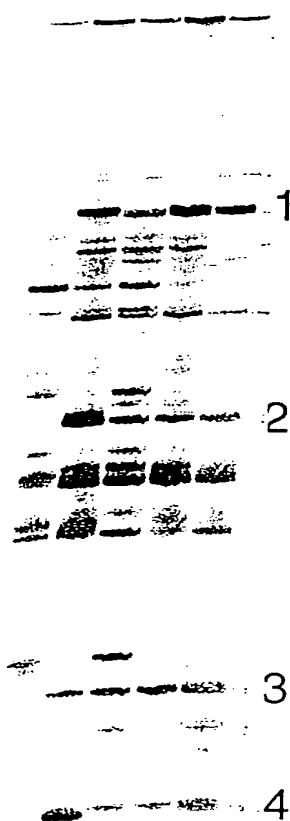
a. B/c.N
b. BCB 13
c. BCA 22
d. BCA 34
e. BCA 39

PCR amplification with
primers MR_1 + MR_5.

Bands 1 to 4 are
examples of
differential display
in tumors.

FIGURE 7

A.

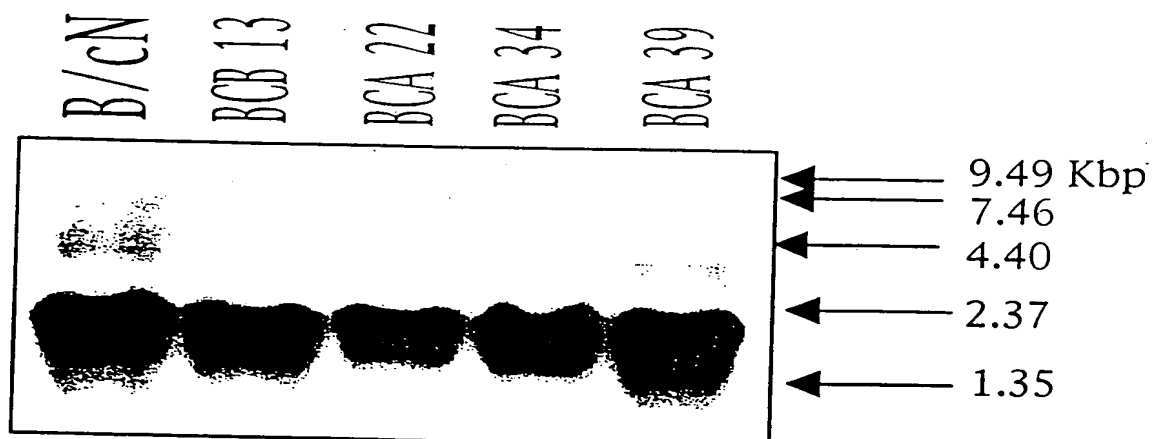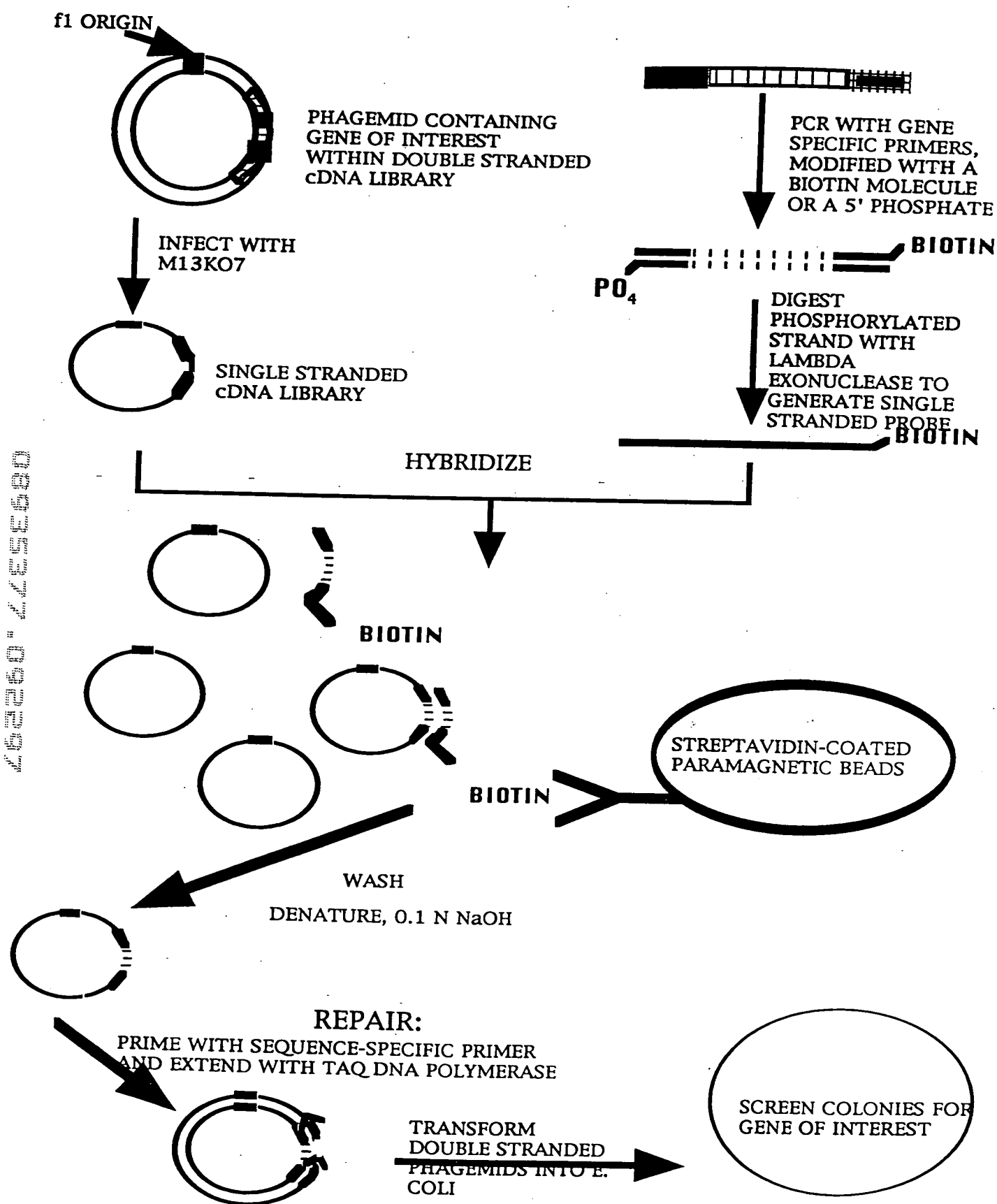
B.

FIGURE 8
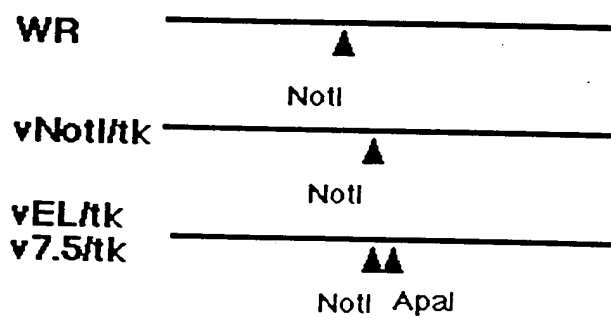
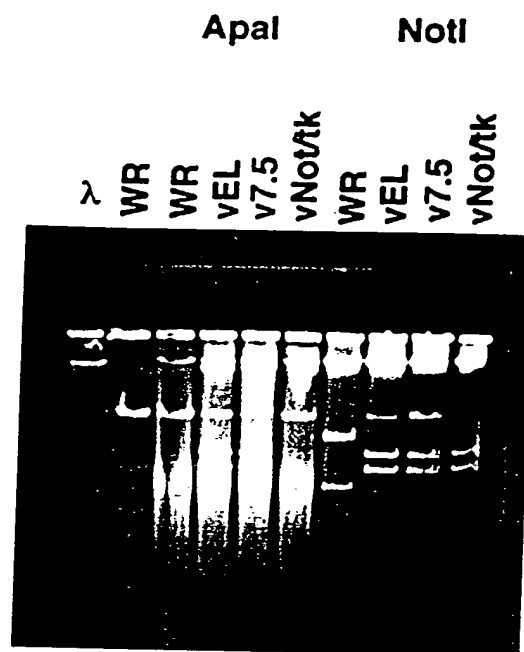f1 ORIGIN

PHAGEMID CONTAINING
GENE OF INTEREST
WITHIN DOUBLE STRANDED
cDNA LIBRARY

PCR WITH GENE
SPECIFIC PRIMERS,
MODIFIED WITH A
BIOTIN MOLECULE
OR A 5' PHOSPHATE

INFECT WITH
M13KO7

BIOTIN

PO_4

SINGLE STRANDED
cDNA LIBRARY

DIGEST
PHOSPHORYLATED
STRAND WITH
LAMBDA
EXONUCLEASE TO
GENERATE SINGLE
STRANDED PROBE

BIOTIN

HYBRIDIZE

BIOTIN

STREPTAVIDIN-COATED
PARAMAGNETIC BEADS

BIOTIN

WASH

DENATURE, 0.1 N NaOH

REPAIR:
PRIME WITH SEQUENCE-SPECIFIC PRIMER
AND EXTEND WITH TAQ DNA POLYMERASE

TRANSFORM
DOUBLE STRANDED
PHAGEMIDS INTO E.
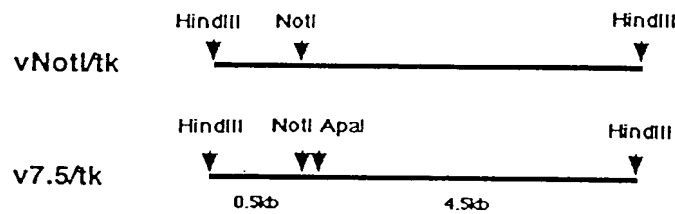COLI

SCREEN COLONIES FOR
GENE OF INTEREST

FIGURE 9

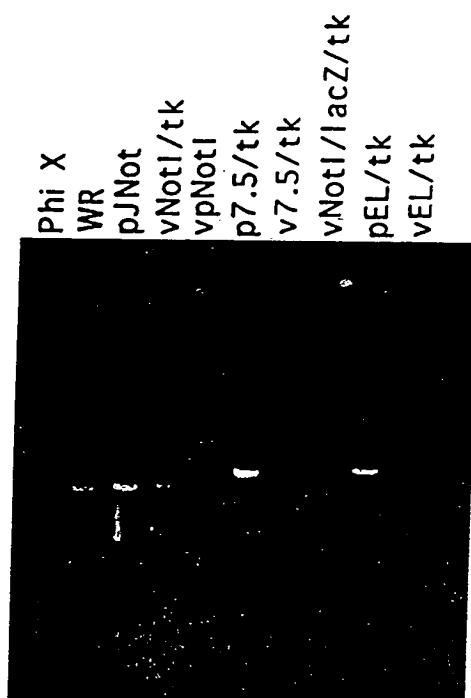FIGURE 10

A

FIGURE 11

B
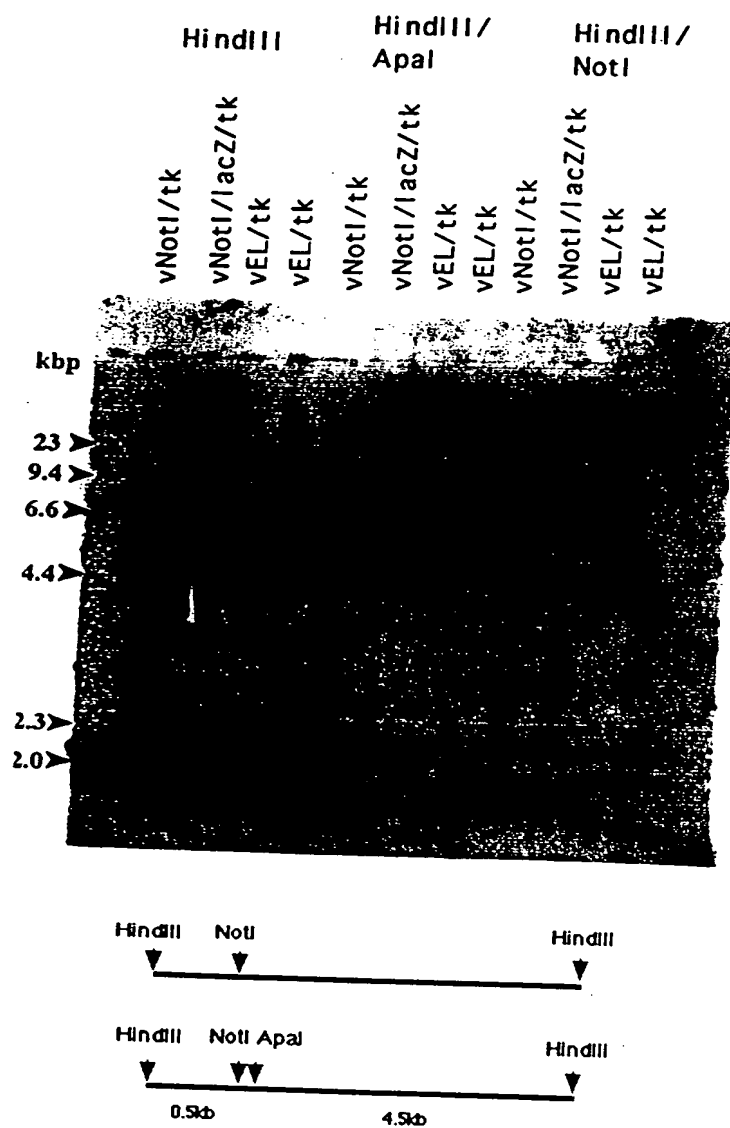
FIGURE 11

FIGURE 12
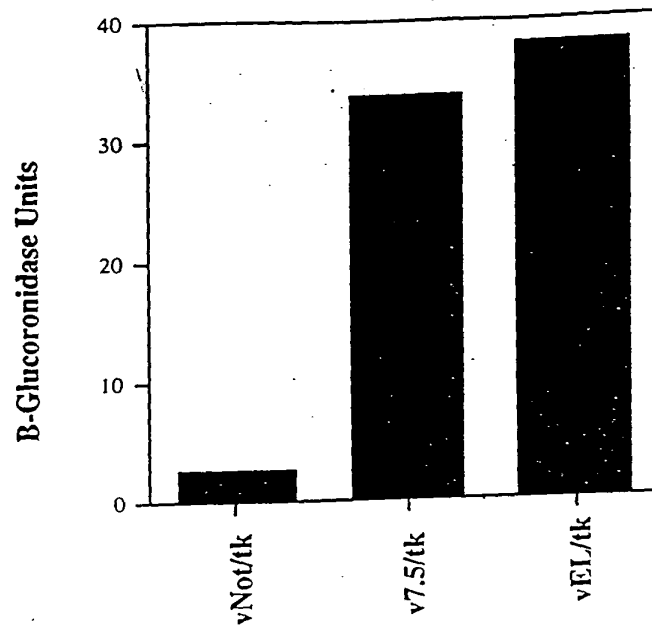
B-Glucoronidase Analysis of Recombinant Viruses

FIGURE 13

FIGURE 14